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STUDY OF TOXIC EFFECT OF MONOCROTOPHOS 36% E.C ON THE BIOCHEMICAL CHANGES IN FRESH WATER FISH *CATLA CATLA* (HAMILTON, 1882)

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ABSTRACT

This study was undertaken to find out the Catla catla fresh water fish biochemical changes in the fish muscles, liver, and kidney. Toxicity were calculated probit analysis, The total carbohydrate contented was estimated by the technique of Roe, Protein was estimated by Lowry method and total lipids were estimated by the method of floch methods. The results shown on total carbohydrate level in muscle 6.43 mg/g after treatment 2.70 mg/g, liver carbohydrate control 17.55mg/g after treatment 16.10 mg/g changes accrued 7.41 % and kidney carbohydrate control 1.44mg/g after treatment 0.53mg/g was gradually decreased. Then total protein in muscles 23.20 mg/g after sublethal concentration at 96 hours treated 17.80mg/g decreased and liver protein 23.12mg/g treated fish 19.40 mg/g changes accrued 13.58% kidney protein level 9.60 mg/g after treatment in sublethal concentration in 96 hours 6.24 mg /g were decreased and Total lipid level are in muscles 1.78mg/g after 96 hours 0.89mg/g liver lipid 7.10 mg/g after treatment 4.35mg/g then kidney 2.09mg/g 0.92 mg/g simultaneous gradually decreased muscles, liver and kidney. The Monocrotophos affects not only fishes but also organisms in the food chain through the procedure of expenditure of one by the other those human beings affected various genetic disorders absolutely insecticides.

Keywords – Monocrotophos, Catla catla, Carbohydrates, Protein, Lipids.

1. INTRODUCTION

Pesticides are generally used in contemporary agriculture to aid in the manufacture of high quality food. However, some pesticides have the probable to cause serious environment and health directly or ultimately¹. Long-term expose function of toxic chemical as well as pesticides in different ecosystems, which due to their high efficacy and easy use have eliminate some biological methods of pest control, was caused by ecological pollution ²Developed and developing countries which are succeeding quickly in the field of agriculture, technology and industry are incessantly release a variety of kinds of harmful substance into the biosphere and in this manner cause a brutal hazard to the environment ³⁻⁴. The most important source of irrigate pollution are household, agricultural and

industrialized waste which are discharge into ordinary water body ⁵. Domestic sewages are sprint decomposed commencement agriculture field weighed down with pesticides and fertilizers contaminate the water body. Frequently worn pesticides can be injurious livelihood organisms, pets, and their surroundings. These dropout frequently enclose an assortment of type of pollutant such as heavy metals, radioactive elements, pesticides, herbicides and sarcastic substance like acids and basis⁶ reuse bring into play of pesticides causes element pollution consequences in the near future health hazard to be alive stock, above all to fish, birds, frogs, and mammals all live animals⁷.

Rhythmic experience to sub-lethal dose of some pesticides can cause physiological and behavioral, biochemical and histological changes in fish getting directly ⁸ that reduce populations, such as leaving behind of nests and broods, decreased imperviousness to disease and greater than earlier than failure to let alone predators ⁹Nowadays modern globalization a assortment of factor poignant formers are using authenticity of pesticide, insecticide, herbicide using agricultural countryside. The pesticide mainly two types Organochlorine, organophosphate and various types of mechanism like pesticide insecticide in topical year monocrotophos is organophosphate using their field of scheming the creepy-crawly pest ⁴⁰. Aquatic water bodies are frequently polluted with a numerous of potentially dangerous substance ¹⁰.

Fishes play an very important accountability in human nourishment fish proteins are well balanced with obligatory amino acids and are analogous to other proteins of animals origin ¹¹.Supplementary fishes contains lipids particularly omega fatty acids from the human nutritious position of inspection, eminent and excellence in attendance in maritime and cultivable fishes. Various toxicity data symbolize for assortment of pesticide such as organophosphate, organochlorine, carbamide and pyrethroid pesticides have been reported for number of fish species noted various approaches and momentous researchers find out like ¹²⁻¹⁸. The enzyme activity slowly but surely level of decreases mercury chloride (Pbcl₃) using pesticide martin ¹⁹. Metabolic activity and physiological activity studies alone do not satisfy the accomplished various toxic pathological circumstances of tissue under toxic stress ¹⁵. The natural physiological accomplishment of an organisms get dispersed on reporting to toxicants, stress, it induces its effect first at cellular or even at molecular level, but ultimately cause physiological, pathological and biochemical modification ²⁰.

The biochemical changes occurrence in the body of the organisms give first indication of stress several investigators have description a quantity of change in biochemical parameter of aquatic organisms due to pesticides reporting ²¹⁻²⁵.Carbohydrates form one of the major source of energy forerunner under any stress circumstance. Total carbohydrate satisfied decreased during the revelation to monocrotophos in the air breathing fish *Anabas scandens* maximum decrease in the brain tissues observed on 21st day ²⁶. The total decrease in carbohydrate level has been noted in the liver and muscle of *Heteropneustes fossilis* showing to Butachlor ^{27,1}. Chlorpyrifos, an organophosphate compound diminish hepatic glycogen level due to inactivation of enzymes concerned in the carbohydrate metabolism in the fresh water fishes, such as *Catla catla*, *Labeo rohita* and *Cirrhinus mrigala*²⁸⁻⁴⁰. Momentous fall in the entire biochemical constituent in all the tissues except glucose prompt to propose that the fish cultured in the aquatic systems earlier to the industrial locations would not have the predictable nutritive value ²⁹. Because of their low insistance, frequent application of these pesticides are life form skillful for the have power over of pests in agricultural fields and thereby large quantities find their way into water bodies.

Protein being the essential substance is desirable for growth and enlargement and also serves as energy source during the stress condition. The total protein point of muscle and liver are decreased in freshwater teleost fish, *Channa punctuates* showing to Nuvacron ³⁰. Decreasing trends have been reported in gill, liver, muscle and brain tissues of *O. mossambicus* exposed to Quinalphos ³¹. Lipids are in general triglycerides that can dish up as metabolic reserves. Phospholipids show a quick diminish given that it is aggressively degraded due to the pesticide stress ³²⁻³³ reported decrease in liver lipid content of *Barbus chonchonius* exposed to Aldiocarb for 15 and 30 days for sublethal concentration.

In general, the end point worn in toxicity studies is mortality, survival and growth with acute toxicity tests, the consideration are quite apposite, but for long-term sublethal concentration's this applicable parameter are easier said than done to ascertain ³⁴. Organophosphate (OP) pesticides are pronouncement increasing use in recent years since they are biodegradable therefore keep it up in the environment only for a short time. Therefore, in the present study enhance and accommodating to such as good finding an the assessment of biochemical components like a total carbohydrates, protein and total lipids, in the muscle liver kidney and studies of the fresh water fish in *Catla catla*. Now day's farmers are using an assortment of pesticide and insecticide monocrotophos in their grassland of cultivation devious the insect pest. Residual of this pesticide alters in to the ecosystem and trouble the healthy environment and aquatic forms. Aquatic farm contains fish and other organism. But the fish is mostly affected by pesticide residuals.

2. MATERIALS AND METHODS

2.1 Acclimatization

Healthy freshwater fish *Catla catla* finger links of the weight ($10 \pm 1g$) and length (8.0 ± 0.5 cm) were selected for the experiment and were collected from the local commercially culture farm Kumbakonam near Swamimalai HAQ fish culture pond. Fish were screen for any pathogenic infections. Glass contamination aquaria were washed with 1% KMnO₄ to keep away from fungal contamination and then dried in the sun light. In good physical shape fishes were then transfer to glass aquaria (35:20:20 cm) containing dechlorinated tap water. Fish were acclimatized to laboratory conditions for 10 to 15 days prior to carrying out tests. The rate of humanity during becoming accustomed was less than 10%. They were habitually fed with marketable food. Chlorinated tap water was represented every day to take not here face and food stuff bits and pieces.

2.2 Toxicity test

Toxicity tests were conduct in harmony with customary methods 3. Accumulation solution of monocrotophos 36% EC with an attentiveness of 0.1 ml per liter (equivalent to 1 ppm) was prepared in distilled water. Based on the progressive bisection of intermission on a logarithmic scale, log concentrations were fixed after conducting the assortment pronouncement test. The fish were starved for 24 hours aforementioned to their use in the experiments as not compulsory by luggage compartment to avoid any intrusion in the toxicity of pesticides by excretory products. After the adding together of the toxicant into the test tank with 10 liters of water have twenty fish, mortality was record after 24, 48, 72 and 96 hours. Five replicates are maintained concurrently. Percent mortality was premeditated and the morals were transfer into probit scale 3.

Probit analysis was carried out as recommended by 34. Deterioration appearances of probit alongside logarithmic transformations of concentrations were made. Off the record limits (upper and lower) of the regression line with chi-square test were calculated by an automated curriculum SPSS Version 14 for 34 Probit analysis.

2.3 Methods

2.3.1 Total carbohydrate estimation

The total carbohydrate at ease was humdrum by the practice of 35. A 10% homogenate of tissue muscles, liver, kidney was equipped using 5% TCA and this was centrifuged at 3000 rpm for 10 minutes. Samples were cooled in the dark at space temperature for 30 minutes. The supernatant was collect and the optical density was deliberate in a spectrophotometer (Hitachi 2205) at a wavelength of 620 nm a blank explanation. Blank was geared up by integration 1 ml of distilled water with 4 ml of Biuret reagent. The total carbohydrate content was intended in mg/g of tissue.

2.3.2 Total protein estimation

Protein was estimated by the method of 36. 1% tissue homogenate were outfitted in 10% TCA and centrifuged at 3000 rpm for 15 minutes. The gal set was liquefy in 1 ml of 1N NaOH to the higher than 5 ml of alkaline copper reagent was added and after 10 minutes,

0.5 ml of folin phenol reagent was considered after was accompanying and quickly The mugginess content was estimated by subtracting the dry heaviness (dried in hot air oven) recognized wet of the tissue muscles, liver, kidney.

2.3.3 Lipid estimation

The total lipids were extract by the method of 37 to find out total lipid, known volume of conduct experiment samples were homogenized with 1 ml of methanol and 2 ml of chloroform to which again 2ml of chloroform : methanol (2:1 v/v) was added and mixed systematically. To this, 0.2 ml- 0.09 % sodium chloride explanation was added. The above mixture was poured into independently funnel, mixed and permissible to situate for few hours.

The lower phase was alienated and 0.5 ml of extract was deliberate and poured into a clean test tube. It was allowed to try in vacuum dedicators over silica gel, dissolve in 0.5 ml concentrated sulphuric acid and mixed well. The tube was plug with non- absorbent cotton wool and placed in a sweltering water bath for 10 minutes and the tubes were cooled at room temperature. 0.3 ml of this acid absorb was taken for experimental analysis. 0.5 mg of cholesterol for stand and, 0.5 ml of distilled water for blank unconnectedly. To each tube, 5ml of vanillin reagent was added. Mixed well and allowable to stand for half an hour and the developed color were deliberate at 250 nm.

3. RESULTS

After sublethal concentration all the way through the path of experiment surveillance were made to approximation how the animal reacts to the toxicity consequence of the pesticide monocrotophos. From the **(Table-1)** it is manifest that the control commonplace carbohydrate in muscle 6.43 mg/g after the exposure period of 24 hrs. The carbohydrate was reduced to 5.83 mg/g liver carbohydrate 7.55 mg/g and 15.40 mg/g 1.42 mg/g respectively for 48, 72, 96 hrs of exposure period. Liver tissues shows **(Fig.-1)**control 17.55 mg/g after 96 hours changes 16.10 mg/g kidney have significant changes be in indict of tissues 1.44 mg/g treatment 0.53 mg/g 37% changes carbohydrate level. Homogenate cell were notice protein have recurrently changes occurs 23.20 mg/g after treatment sublethal concentration 17.80 mg/g from muscles cells. The results shows **(Table: 2)** Then liver cells 23.12 mg/g and changes **(Fig: 2)** accuracy 19.40 mg /g protein level were slowly but surely decrease kidney control protein shows were 9.60 mg/g after 96 hours total protein level was decrease 32.39% protein amalgamation have been accrued total protein level decreased in highest level because the metabolic activity and urinary waste was remove therefore total protein level decreases in kidney tissues.

The body alteration and synthesis problem regulation in present of lipids are total values gradually decreased **(Fig:3)** lipids level control *Catla catla* fish 1.78 mg/g muscles and changes 0.89 mg/g liver tissues gradually decreased on 4.34 mg/g then kidney tissues control 2.09 mg/g after 96 hours sublethal concentration 0.92 lipid level was decreased after 48 hour highest level in kidney 28 % changes **(Table : 3)**. The total carbohydrate, protein, lipids and various tissues like Muscles, liver and kidney gradually decreased shows on carbohydrate, protein and lipids.

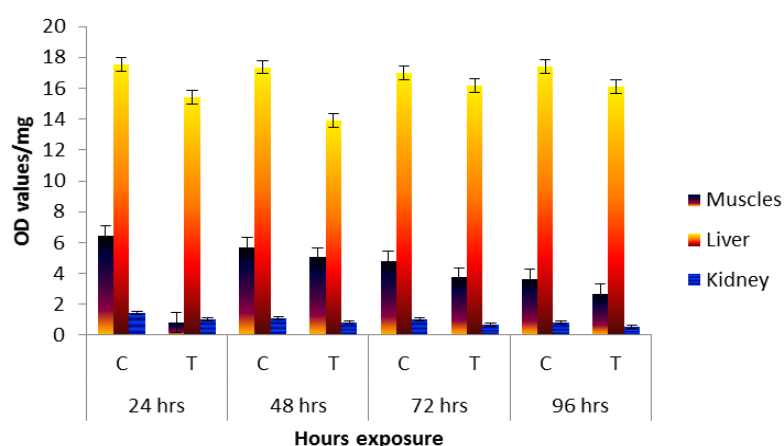


Fig 1. Error Diagram Hours of exposure Carbohydrate level in *Catla catla* fish against monocrotophos

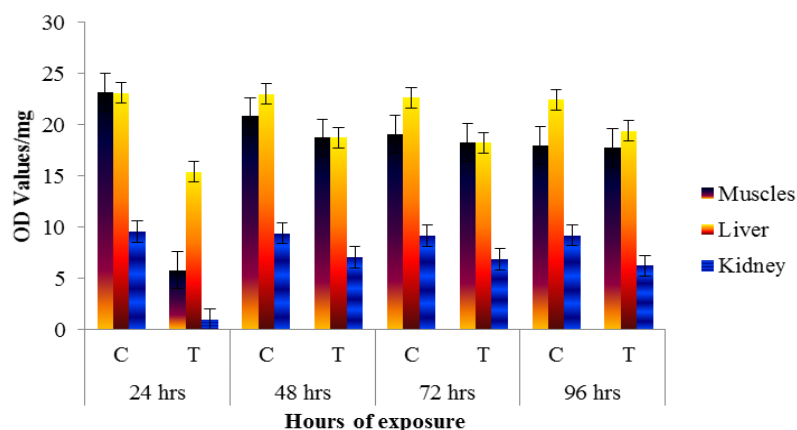


Fig 2. Error Diagram Hours of exposure Proteinlevel in *Catla catla* fish against monocrotophos

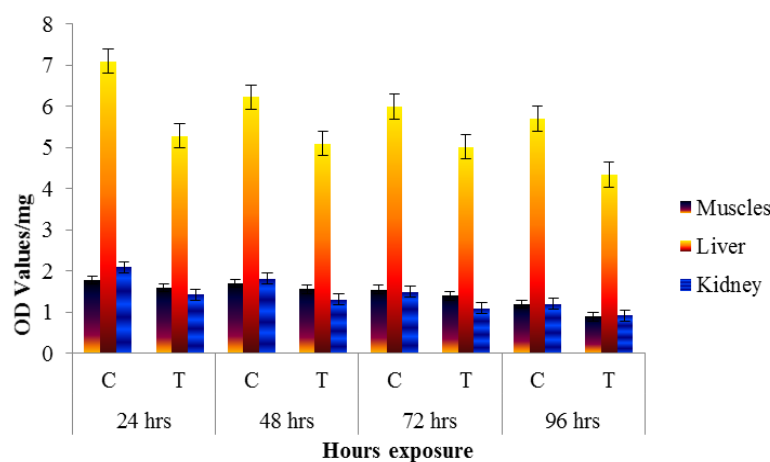


Fig 3. Error Diagram Hours of exposure Lipidslevel in *Catla catla* fish against monocrotophos

Table 1: Hours of exposure Carbohydrate level in *Catla catla* fish against monocrotophos

Test	Tissues	24 Hours			48 Hours			72 Hours			96 Hours		
		C	T	CH %	C	T	CH %	C	T	CH%	C	T	CH%
Carbohydrate	Muscles	6.43±0.01	5.83±0.21	9.33	5.72±0.03	5.04±0.01	13.49	4.80±0.021	3.75±0.12	21.87	3.66±0.16	2.70±0.01	35.55
	Liver	17.55±0.06	15.40±0.03	12.25	17.37±0.01	13.88±0.01	20.09	17.00±0.03	16.20±0.01	4.70	17.39±0.12	16.10±0.16	7.41
	Kidney	1.44±0.20	1.02±0.07	0.29	1.12±0.06	0.85±0.03	24.10	1.00±0.01	0.66±0.01	0.34	0.85±0.21	0.53±0.18	37.64

Value are Mean ± SD of six observation – or + indicate percentage decrease or increase over control. (n=6)

Table 2: Hours of exposure protein level in *Catla catla* fish against monocrotophos

Test	Tissues	24 Hours			48 Hours			72 Hours			96 Hours		
		C	T	CH %	C	T	CH %	C	T	CH%	C	T	CH%
Protein	Muscles	23.20±0.12	21.20±0.04	0.86	20.87±0.01	18.75±0.09	10.15	19.10±0.08	18.30±0.11	4.18	18.00±0.01	17.80±0.03	1.11
	Liver	23.12±0.08	18.83±0.01	0.18	23.00±0.02	18.78±0.03	18.34	22.66±0.11	18.25±0.01	19.46	22.45±0.06	19.40±0.07	13.58
	Kidney	9.60±0.01	7.24±0.02	0.24	9.42±0.18	7.10±0.10	24.62	9.20±0.18	6.90±0.01	0.25	9.23±0.02	6.24±0.01	32.39

Value are Mean ± SD of six observation – or + indicate percentage decrease or increase over control. (n=6)

Table 3: Hours of exposure lipids level in *Catla catla* fish against monocrotophos

Test	Tissues	24 Hours			48 Hours			72 Hours			96 Hours		
		C	T	CH %	C	T	CH %	C	T	CH%	C	T	CH%
Lipids	Muscles	1.78±0.06	1.60±0.01	0.13	1.70±0.23	1.56±0.06	8.23	1.55±0.02	1.40±0.06	9.67	1.20±0.01	0.89±0.03	25.83
	Liver	7.10±0.02	5.29±0.10	0.25	6.23±0.01	5.10±0.01	18.13	6.00±0.06	5.02±0.07	16.33	5.70±0.01	4.35±0.01	24.21
	Kidney	2.09±0.05	1.43±0.01	0.31	1.82±0.01	1.31±0.04	28.02	1.50±0.19	1.10±0.02	26.66	1.20±0.03	0.92±0.01	23.33

Value are Mean ± SD of six observation – or + indicate percentage decrease or increase over control. (n=6)

4. DISCUSSION

The ever-increasing use of pesticides causes chemical pollution results potential health hazards to live stock, in particular to fish, frogs, birds and mammals³⁸. The physiological movements in animal were effected with the aid of muscle. The following are the principal constituents of muscle: water 75%, protein 20%, minerals and organic compounds 5%. Proteins are perhaps the most essential and typical of all the constitutions of livelihood cells¹. Proteins constitute the fabric material of protoplasm. Apart from formation of protoplasm, protein is an important constituent of the various cellular membranes in conjugation with lipids. Most of the biological active compounds are proteins including enzymes¹². The use of protein fuels is inadequate for the reason that they cannot be store like lipids and carbohydrates.

Proteins are complex substance with high molecular compound weight form not only the structural framework, but also gears and levers of the operating mechanism in the living wage body. Proteins are useful for the polypeptide chains of amino acid molecules²⁷. The proteins are useful for the transport and storage. Specific proteins transport many small molecule and ions. A protein was complex that guides the formation of neural networks in higher organisms. The primary function of protein food is to supply the amino acids needed for the growth, repair and general maintenance of the structural and catalytic machineries of living.

Carbohydrate and protein are the chief nutrients of the animals. They have a variety of function. The carbohydrate supplies energy in the form of ATP molecules, which are formed during TCA cycle. The proteins in different tissues differ in composition and properties¹⁹. In the present study the protein content in the muscle and liver kidney of *Catla catla* is decreased with the low concentration of pesticide Monocrotophos. Even with the same concentration longer exposure resulted in decreased amount of protein content (**Table: 1**) which indicate that the tissue protein endure proteolysis. The result in the production of free amino acids, which are used in TCA cycle for energy production under, stresses³⁷. There are similar reports of effects of toxicants on total protein in other fishes by^{6-7, 39}.

In the present study the result obtains clearly indicate that there was a decreased amount of protein and glycogen content to resist the effect of pesticides. That is to provide immediately energy to the combating elements of the body and protect all systems of the body from the harmful effect of the pesticide¹⁸. With regard to carbohydrate in the fish exposed to different hours of exposure of pesticide Monocrotophos there was no much change within 24 hours of treatment with low concentration 0.040 ppm⁴⁰ fish kept prolonged exposure up to 96 hours the carbohydrate content was observed in decreased amount (**Table : 1**) the carbohydrate content was found more and more in decreased. Such reduction in store carbohydrate content has been reported in *Labeo rohita* exposed to monocrotophos¹⁹⁻²⁰ effect of monocrotophos was reported by³². A fall in carbohydrate levels clearly indicates its rapid utilization to meet the enhanced energy demands in pesticides treated individuals through glycolysis or hexose monophosphate pathway⁶.

The term lipid was used by the biochemist to describe that group of substances of animal origin, which is inexplicable in water, but soluble in fat solvents¹⁷. All cells contain lipid in the form of globules scattered in the cytoplasm. The concentration is much higher in cells forming adipose tissue. In the present study hours of exposure periods has decreased amount of lipid. Totally concluded pesticide

posses through high health impact entire food chains various human disorders attains various aspects in human body then avoid pesticide using biopesticides.

5. CONCLUSION

The results in the present study showed that the various hours of exposure in *Catla catla* fresh water fish against monocrotophos caused by various physiological changes and pathology in their organs muscles liver and kidney they were associated with various cell signals and metabolic activity in the exposure. It can be used as a sensitive model to monitor the aquatic pollution, aquatic animals and using organophosphate various via human genetic disorders in young one generation.

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8. CONFLICT OF INTERESTS

The authors declare that there is no conflict of interests regarding the publication of this paper.

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